IN[redacted]E UNITED STATES PATENT AN[redacted]ADEMARK OFFICE

APPLICANT	:	SAMUEL ROSE, MD
SERIAL NO.	:	08/782,590
FILED	:	January 13, 1997
FOR	:	A METHOD AND COMPOSITION FOR TREATING CANCER BY CONVERTING SOLUBLE RADIOACTIVE TOXIC AGENTS INTO INSOLUBLE RADIOACTIVE TOXIC PRECIPITATES VIA THE ACTION OF NON-MAMMALIAN ENZYMES BOUND TO THE NON-ENDOCYTOSING RECEPTORS OF CANCER CELLS
EXAMINER	:	SUSAN UNGAR, PH.D.
GROUP ART UNIT	:	1640

Commissioner of Patents and Trademarks
Washington, D.C. 20231

SIR:

1. I, retired Professor of Immunology at Roswell Park Cancer Institute, George L. Mayers, hereby declare that I am a citizen of the United States and a resident of Gainesville, Florida.

2. I am currently retired Professor of Immunology from Roswell Park Cancer Institute. I have been in the field of cancer research for over 30 years, was former Chairman of the

Department of Immunology at Roswell Park, and served as a Professor and Chairman of the Department of Immunology and Microbiology, Roswell Park Division, State University of New York at Buffalo. Principally throughout my career I was a working research scientist in the field of cancer biology and cancer therapy.

3. I make this declaration under 37 C.F.R. 1.132 to traverse grounds of rejection of the above-identified U.S. Patent Application, Serial No. 08/782,590 of Samuel Rose, MD (hereinafter the '590 Application).

4. I have examined and am familiar with the original specification, claims, and drawing of the '590 Application; the Official Action mail March 18, 1998; the Amendment filed September 21, 1998, the Official Action Mailed November 25, 1998, the Official Advisory Action August 16, 1999, the Preliminary Amendment for Continued Prosecution Application November 16, 1999, and the Official Action, May 25, 2000. I have also examined and am familiar with the excerpts from 2 confidential, unpublished reports, dated January 17, 1997 and June 20, 2000, prepared by Marin Biologic Laboratories, Inc.(a contract research laboratory) which provide in vitro scientific data supporting the present invention.

REGARDING OFFICIAL ACTION MAILED MAY 22, 2000, PAPER NUMBER 27 (HEREINAFTER "ACTION")

5. **Regarding Action, page 4 (line 4-12), page 4 (line 14-17), page 6 (line 15-17), page 6 (line 20-22), page 8 (line 3-5), and page 8-9 (line 21- line 2) – (Response 1)**

In these sections and others, the Examiner raises concerns over whether the first therapeutic agent claimed in the '590 Application will function as disclosed, whether accurate predictions can be made regarding the function of the first therapeutic agent

such that the present invention can be practiced. The method of the present invention is an improvement and evolution of the prior art in the field of cancer therapy known as Antibody Dependent Pro-Drug Therapy (ADEPT) which was documented by Dr. Rose in the specification of the '590 Application on pages 9-10. I think that a general problem may arise from a misunderstanding of the definition of terms concerning the use of the term therapeutic agent as read by the examiner and used by the applicant. The formation of the whole complex provides the therapy by laying down a precipitated radiolabeled reagent within the tumor, which led to calling the several components therapeutic agents. However, many of the initial components that are termed therapeutic agents in this scheme are not in and of themselves capable of providing any therapeutic effect. It is only when all the therapeutic agents are in place that the addition of the final high specific activity radiolabel that is induced to precipitate within the tumor that a therapeutic effect can be achieved. In the present invention the first therapeutic agent is converted by the enzyme moiety of the first bispecific agent into the first extracellular precipitate, which is insoluble. The specification of the present invention enables one of skill in the art to predict that the first therapeutic agent will function as claimed and specifically that it will precipitate via enzyme action of the first bispecific reagent. One skilled in the art would recognize that enzymes could be used to catalyze a wide variety of chemical reactions, which would not occur without the enzyme being present. Indoxyl derivatives have received extensive use to visually identify cellular structures targeted by antibody-enzyme conjugates by the formation of indigoid precipitates. Holt, et al. has extensively studied this area to show that precipitates can be achieved with almost no diffusion so that the fine structure of cellular components can be clearly identified by the antigenic determinants bound by the antibody (Proc. Roy. Soc. B. **142**, 160, 1954; Proc. Roy. Soc. B. **148**, 506, 1958; Proc. Roy. Soc. B. **148**, 495, 1958; Proc. Roy. Soc. B. **148**, 520, 1958). Many others have more recently used another component of the oxidation of indoxyl to indigo; i.e., the release of hydrogen peroxide to be used as a substrate in many different chemifluorescence assay systems. This patent makes use of the action of enzymes on indoxyl compounds in a new way to produce an indigoid precipitate, which will act as a platform or scaffolding with new epitopic sites that provide positions at which to carry out therapeutic reactions. This is clearly different from ADEPT where the

maximum number of enzyme molecules is limited by the number of useable epitopes present on the surface of tumor cells. The formation of the scaffolding or platform that is compatible with relatively long term host existence allows for much greater levels of amplification to occur before administering the final therapeutic agent capable of locally irradiating the tumor and eliminating it. Thus given the disclosure in the specification of the '590 Application and given the widely published literature on the field of ADEPT and indigo assays, one of skill in the art would be able to predict that the first therapeutic agent would function as claimed and one would be able to practice the present invention without undue experimentation.

which is a valid model (Supported January 17, 1997 from Marin Biologic Research disclosed and that the function of the first therapeutic agent can be predicted despite the absence of in vivo data because the first therapeutic agent precipitated in serum as a function of the physically supported enzyme and the precipitate formed in the immediate surroundings of the localized enzyme. The serum provides and environment that contains the in vivo constituents around the beads with the enzyme that would be expected if it were localized in the tumor.

Regarding Action, page 4-5 (line 19-2) – (Response 3)

To one of skill in the art it is evident that the only location where precipitation will occur will be at the site of the enzyme moiety of the first bispecific reagent. As discussed above, enzymes catalyze reactions, which would not occur if the enzyme were not present. The function of the indoxyl variety of the first therapeutic agent described in the present invention is an example of such an enzyme catalytic reaction. As described in the specification of the '590 Application and extensively published in the field of indoxyl chemistry (both in the field of indigo dyes and more recent applications using indoxyls as part of chemical assays) precipitation via enzyme action occurs instantly – once the material at position 3 of the indoxyl is cleaved by the enzyme, the indoxyl dimerizes and precipitates. This has been extensively studied and verified by the work of Holt et al. mentioned above. Many of these indoxyl derivatives can be commercially purchased

(see for exa[redacted]e the Sigma or Inalco catalogs), thus sh[redacted]ng that they are stable and do not precipitate without the action of the appropriate enzyme. In addition, the in vitro studies documented in the June 20, 2000 report from Marin Biologic Laboratories, Inc. have shown that a number of these compounds (13 different indoxyl derivatives) have been evaluated in human serum for 4 days for formation of indigo in the presence of the physiologic enzymes present in serum (which also closely reflects the enzymes present in the extracellular fluid) and shown that at the end of 4 days all but indoxyl phosphate formed less than 2 micrograms of indigo from 1 mg of indoxyl derivative; indoxyl phosphate produced 10 micrograms of indigo from 1 mg of indoxyl phosphate in 4 days. The experiments show that there are essentially no enzymes present in the extracellular fluids that convert indoxyls to indigo and that it does not happen spontaneously in the extracellular fluids.

Regarding Action, page 5 (line 6-10) – (Response 4)

As discussed in the Declaration of Dr. Alan Epstein submitted previously and in the specification of the '590 Application, published research in the field of tumor biology describes how tumors are known to have very low flow rates (some of these references have been cited in the specification of the present invention on pages 35-36, in Exhibits B and C submitted with the response filed May 25, 1999, some have been cited by the Examiner, and others are well-known in the field). Further, it is known that in developed tumors, there is very dense packing of tumor cells. Thus, even in the absence of data, one of skill in the art would predict that the precipitate formed in the extracellular space of the tumor will remain trapped between the cells and that the precipitate would be removed very slowly. This is also supported by the Marin Biologi data on the beads because examination of the ebads showed trapped indigo precipitate caught in the matricies of the beads. In addition, as described in the specification (page 35-36) and as known to one of skill in the art, not only do tumors have a very slow flow rate compared to normal tissue, but tumors also lack macrophages, lymphocytes, and monocytes and thus precipitate formed in the extracellular space of the tumor would not be removed as quickly as from normal tissues where macrophages, monocytes and lymphocytes are active and rapidly remove unwanted particles. Many histological studies of tumors have shown large

numbers of macrophages and lymphocytes in the tissue volume surrounding a tumor, but extremely few, if any, within the tumor volume. This is an observation that has not been explained. However, when tumors undergo remission, macrophages do enter the necrotic areas and help in the clean up. Oncologists as far back as the 1970s have tried to activate the macrophages surrounding the tumors as a form of therapy without much success. Based on the slower, tortured flow rates known to occur in tumors and the absence of macrophages and lymphocytes within the tumor to help remove insoluble material, one of skill in the art could reliably predict that the rate of removal of precipitate from the tumor would be very much slower from tumor tissue compared to normal tissue.

CONCLUSION

In conclusion, I believe that the specification and claims of the present invention enable one of skill in the art to practice the invention without undue experimentation. I am confident of this conclusion because, given the information presented by Rose in the specification of the present invention (including the numerous references), the information in the public domain related to the claimed invention, and the in vitro data in the reports prepared by Marin Biologic, one of skill in the art can successfully predict (a) that the first therapeutic agent will function in vivo as described, (b) that the only site of conversion of the first therapeutic agent into the first extracellular precipitate will be at the enzyme moiety of the first bispecific reagent, (c)that the therapeutic data published in the field of ADEPT and published data in the field of radioisotope-based therapies (like the treatment of thyroid cancer with radioiodide) enable one of skill in the art to practice the present invention, including doses and methods of administration for all of the claimed reagents, without undue experimentation, (d) that the publications cited by Rose in the '590 application, the references submitted to the Examiner from previous responses, and the general knowledge of the field of tumor biology teach that insoluble materials in the tumor tissue will be removed much more slowly than normal tissue and thus that the insoluble precipitates formed in the present invention will remain in the tumor tissue and will function as claimed.

6. I hereby [redacted]are that all statements made herein of [redacted]wn knowledg are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine, imprisonment, or both, under Section 1001 of Title 18 of the United States Code.

Date: 27 Nov 2000 Signed: [signature]